United States Department of Agriculture

Progress Report

Title:	**Ecobiology, Impact, and Management of Grapevine Red Blotch Virus and its Vector(s) in California and Oregon Vineyards**		
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Non-Technical Summary

Grapevine red blotch disease, caused by the virus Grapevine red blotch virus (GRBV) is an urgent problem for the $162 billion US grape industry. GRBV is a prominent disease found in the majority of grape growing regions in California and Oregon. The grape industry currently lacks best practices for detecting and preventing spread of GRBV within and among vineyards. The discovery of S. festinus as a vector of GRBV significantly increased the possibility of better understanding the epidemiology of GRBD and ultimately its management. However, GRBD spread also occurs in vineyards where S. festinus has not been found. Therefore, information on potential additional vector species in these regions is paramount. Replanted vineyards in California and Oregon have experienced reinfections and a better understanding on the prevalence of GRBV and assessment of risk factors are needed. Proposed research will address knowledge gaps involving the epidemiology of the virus as driven by studies on its vectors and determining how the disease affects grapevine performance and grape quality. The economic impact of GRBV infection on producers and nurseries will also be determined. Sustainable GRBV management strategies developed from the project will be implemented to enhance economic and social impacts and to reduce the impact on environment. This project brings together researchers, extension specialists and stakeholders from CA and OR to help solve a significant new problem facing this valuable specialty crops industry. Outreach activities will be extended to the other states and can thus impact the grape industry in the country.

Accomplishments

Accession No. 1020210 **Project No. CALN-SW-ADM-7934-OG**

Major goals of the project

Grapevine red blotch disease, caused by the virus Grapevine red blotch virus (GRBV) is an urgent problem for the $162 billion US grape industry. This proposal aims to bridge knowledge gaps while generating strategies to manage GRBV. We intend to determine virus presence and spread in states across the US, and baseline knowledge on the risk of GRBV to wine grape production in states where acreage is increasing. Research includes studies on the role of its vector(s) in GRBV spread; economic assessment of GRBV impact on grape production, wine quality, and nurseries; as well as identification of sustainable management options. The latter will have the dual goals of reducing its spread across the US and determine its economic impact on all facets of the wine grape industry, including growers, nurseries, wineries, and state and federal regulatory agencies.
Objective 1. Assess the prevalence and rate of spread of GRBV and the impact of red blotch disease on production and fruit quality in US grape production areas.
Sub-objective 1.1 Determine prevalence and spread of GRBV in US grape production areas.
Subobjective 1.2 Identify alternative hosts and reservoirs of GRBV.
Subobjective 1.3 Establish the effect of GRBV infection on grape, juice and wine quality.
Subobjective 1.4 Assess economic impact of red blotch disease on wine and grape production and nurseries.
Objective 2. Identify and develop sustainable strategies to manage GRBV and its vector(s).
Subobjective 2.1 Determine presence and biology of potential treehopper vectors in symptomatic vineyards and associated crop and non-crop landscapes.
Subobjective 2.2 Improve knowledge of GRBV acquisition and transmission by its vector(s).
Subobjective 2.3 Identify sustainable IPM strategies for the management of GRBV vector(s).
Subobjective 2.4 Identify effective and economically viable strategies to eliminate GRBV sources (infected vines, vineyards, nurseries and landscape plants).
Subobjective 2.5 Evaluate viticultural practices to mitigate the impact of GRBV on infected grapevines in relation to fruit production and quality.
Subobjective 2.6 Develop cost-effective GRBV diagnostic tools - a prototype device to detect sub-visible changes in grape leaves at early stages of GRBV infection.
Subobjective 2.7 Development of a field level detection method based on isothermal amplification.
Objective 3. Implement and evaluate grapevine red blotch disease management programs.
Subobjective 3.1 Evaluate grape industry knowledge of red blotch disease and management.
Subobjective 3.2 2 Provide stakeholders with results, management guidelines and other outcomes of the Grapevine red blotch disease SCRI project activities.

What was accomplished under these goals?

Obj.1: Assess prevalence, rate of spread, and impact of Grapevine red blotch virus (GRBV) on production and fruit quality in US wine grape production areas.
Subobj. 1.1: We have identified vineyards in California's Napa County and Sierra Foothills, and in Oregon's Willamette Valley and southern Oregon where grapevine red blotch disease (GRBD) is prevalent and patterns of GRBV is consistent with being spread by a vector that will serve as study sites for this project. Depending on vineyard, the number of vines surveyed ranged from 1,000 to 12,000 vines. Disease incidence ranged from 0.6 to 58.8% based on GRBD foliar symptoms at harvest, however, 100 leaf tissue samples were collected from each vineyard to confirm symptom-based observations. We will expand the surveys to more vineyards in 2020-2021 and repeat the surveys and symptom mapping for each vineyard for two to three additional years to determine the spread of GRBV within the vineyards.
Subobj. 1.2: Activities started in Oct. 2020.
Subobj.1.3: Three vineyard sites were identified in Napa and San Luis Obispo counties. A subset of vines was tested by qPCR and GRBV pos and neg data vines identified. Berry chemistry at harvest and yield data per vine were recorded. Wines were made from these vineyards as well as from subobj. 2.5 vine trials at Oakville Exp. Station. Winemaking is ongoing and chemical analysis and sensory evaluations will start in 2021.
Subobj.1.4: Start in 2021.
Obj.2: Identify and develop sustainable strategies to manage GRBV and its vector(s).
Subobj. 2.1: Detailed studies were conducted on treehopper species in California and Oregon and leafhopper species in California. We determined seasonal phenology, distribution, and alternative hosts of Spissistilus festinus, Tortililus wikhami and T. albidosparsus and the leafhopper species Schaphytopius spp. It is clear that the treehoppers were not able to complete their lifecycle on cover crop Brassica spp. (Wild Mustard), but were able to complete their lifecycle on a range of other hosts, including apple, pear, oak, sorghum, pea, several grass species, and vetch. Generally, treehoppers were found mostly on vineyard edges, compared to the Schapytopius spp., which was found in relatively large numbers within the vineyard canopy.
Subobj. 2.2: Controlled greenhouse and laboratory trials were conducted to determine the ability of treehopper populations to

transmit GRBV. Collections of live S. festinus (Say), Stictocephala basalis (Walker), and T. albidosparsus (Stål) (Hemiptera: Membracidae) were made in proximity to commercial vineyards. One male Sp. festinus successfully transmitted GRBV, as determined by quantitative PCR (qPCR) and droplet digital PCR, to a single vine. Testing of individual insects using qPCR revealed that GRBV can persist inside the bodies of all three treehopper species; however, results to date showed no evidence following successive years of testing that GRBV can be transmitted by source populations of St. basalis or T. albidosparsus. Vector acquisition and inoculation of the GRBV causal agent is also being evaluated using field vines for acquisition and potted vines for inoculation. We have successfully shown S. festinus or Scaphytopius spp. on virus-infested vines have been positive for GRBV (e.g., acquisition). However, we have not yet observed transmission to clean vines at UC Berkeley for S. festinus and we are currently holding and testing vines for Scaphytopius.
Subobj 2.3: We validated and published a phenology model to assist prediction of timing to initiate mowing to eliminate vineyard floor plant hosts of S. festinus in late winter and spring using field data collected in winter and spring 2020 at 3 northern CA commercial vineyard locations. A replicated experimental vineyard block was planted in southern OR that will enable comparison of clean cultivation to mowing vegetated vine row middles. The mowed plots were planted with a mix of grass and trefoil, and the groundcover is in the process of becoming established. Unmanaged orchard floor vegetation may serve as an overwintering reservoir for GRBV vectors in western US vineyards.
We also developed and field-tested a substrate vibrational trap during 2020. The first prototypes were trialed in OR during 2020. Part of this work was characterization of unique vibrational signals produced by treehoppers. We successfully trapped T. albidosparsus in Northern Willamette Valley vineyards using the first prototype of the trap. Of particular importance is the adaptability of the trap and vibrational signals can be adapted to produce recorded vibrational signals of both treehoppers and leafhoppers. These traps can be used both for trapping as well as pest management.
Subobj.2.4: Will start in 2021.
Subobj.2.5: Three replicated trials testing various cultural practices were initiated in May 2020 on both healthy (GRBV-) and infected (GRBV+) vines in a commercial Pinot noir vineyard in OR and a Cabernet Sauvignon vineyard in CA. Experiments included manipulating vine water status (control and water deficit), crop load (100% and 30%), and vine nutrition. Experimental vines were confirmed for virus status during dormancy (2019-20), and experiments were laid out in a RCB design with 4-8 single-vine replicates. Yield and yield components (i.e. cluster number, berry number) were determined to primary and secondary metabolite analyses which are ongoing.
In addition to agronomic data collected at harvest, leaf gas exchange, leaf nonstructural carbohydrates, leaf symptom development, and berry development were monitored regularly in selected GRBV- and GRBV+ vines throughout the growing season. Preliminary data indicate that restricting irrigation does not alleviate GRBV impact. Crop removal experiments revealed that carbon starvation is mediated by transient decreases in leaf photosynthesis and is not enough to overcome the effects of GRBD in grapevines.
Subobj.2.6: To establish descriptors for prediction of early-stage red blotch infection, physicochemical changes of early-stage red blotch infected grape vine leaves were evaluated using FTIR, RGB color microscope and multispectral imaging technique. In addition, data for changes in anthocyanin, pectin and total phenolics were acquired. Currently we are analyzing data using PCA, regression models and machine learning models such as light GBM as a predictive model.
Subobj. 2.7: A vineyard in Southern OR with history of GRBD has been identified. Nearly 300 vines were tested for GRBV in 2019 using PCR assay. In 2020, Loop Mediated Isothermal Amplification developed by Romero et al. (2019) were tested to understand field level detection ability of LAMP assays at different phenological stages. The samples collected at these stages from three canopy layers (base, middle, and top of canopy) will be compared and validated with PCR, qPCR, droplet digital PCR (ddPCR), and symptoms data.
Obj.3: Implement and evaluate grapevine red blotch disease management programs.
Subobj.3.1: Surveys of California and Oregon wine industry knowledge on GRBV will be completed by end of year 2.
Subobj.3.2: In OR an in-person all-day GRBV workshop was held in Salem, OR in Nov. 2019 that was attended by more than 60 industry members. In CA three in-person presentations were given at different symposia in Feb. and Mar. 2020. In fall 2020, OSU researchers organized a virtual GRBV webinar series (due to COVID-19 restrictions on in-person meetings). As of this reporting, three webinars have been held with an average of ~120 viewers each, and seven more are planned into Dec. 2020. Similarly, CA held a 'Grapevine Red Blotch Virus' webinar on May 5, 2020 reaching 68 people live with a similar amount of people viewing the posted webinar later.

What opportunities for training and professional development has the project provided?

In the first year of this project three undergraduate and five graduate students as well as six post-doctorates have been trained in the scientific methodology needed to achieve the goals of this project. This ranges from insect to plant sampling, viral analysis, plant physiological measurements, biochemistry, chemistry, and winemaking. New technology is also being investigated for disease detections and more.

How have the results been disseminated to communities of interest?

In-person seminars and workshops prior to March 2020 as well as scheduled zoom meetings to reach stakeholders during the pandemic.
Anita Oberholster, Kaan Kurtural, Frank Zalom, and Mysore Sudarshana presented a UC Davis Dept. of Enology and Viticulture 'Office Hours' webinar on 'Grapevine Red Blotch Virus' on May 5, 2020.
Anita Oberholster also presented at the Current Wine and Winegrape Symposium on February 11, 2020.
Co-PI Kent Daane did two presentations as listed below:
Current knowledge of grape red blotch associated virus vectors. NCPN-Grapes Tier II Committee Meeting. Davis, CA. Feb. 2020.
Trying to understand the spread and control of "Red Leaf" in vineyards. Central Coast Wine Grape Seminar. Salinas, CA. Mar. 2020.
In Oregon, an in-person all-day GRBV workshop was held in Salem, OR in Nov. 2019 that was attended by more than 60 industry members representing all phases of wine grape production. Seminars covered all aspects of GRBD including effects on grapevine physiology and potential cultural management strategies, entomology and potential pest management strategies, virology and diagnostics, and wine production and sensory. In fall 2020, OSU researchers organized a virtual GRBV webinar series (due to COVID-19 restrictions on in-person meetings), covering similar topic areas as in 2019. As of this reporting, three webinars have been held with an average of ~120 viewers each, and seven more are planned into Dec. 2020.

What do you plan to do during the next reporting period to accomplish the goals?

Vineyard sites identified and used during the 2019-2020 funding cycle will be investigated further to identify potential vectors, host plants and study insect biology. Furthermore, the synergistic impact of environment (climate) and GRBV impacts will be studied. Viticultural practices that showed promise in GRBV mitigation will be expanded upon. Furthermore, transmission studies with potential vectors will continue. Understanding the life cycle and biology of potential vectors are key to the development of successful management strategies. In the second year, cost-effective diagnostic tools for GRBV detection will be explored and economic analysis of GRBV impact on grape and wine quality (value) will commence.

Participants

Actual FTE's for this Reporting Period

Role	Non-Students or faculty	Students with Staffing Roles			Computed Total by Role
		Undergraduate	Graduate	Post-Doctorate	
Scientist	1	1.5	2.3	2.5	7.3
Professional	0.3	0	0	0	0.3
Technical	0.5	0	0	0	0.5
Administrative	0	0	0	0	0
Other	0.4	0	0	0	0.4
Computed Total	2.2	1.5	2.3	2.5	8.5

Student Count by Classification of Instructional Programs (CIP) Code

Undergraduate	Graduate	Post-Doctorate	CIP Code
1	2	4	26.07 Zoology/Animal Biology.
		1	01.11 Plant Sciences.
2	3	1	01.10 Food Science and Technology.

Target Audience

The project has completed its first year of funding and limited outreach and extension have been undertaken due to preliminary data generated as well as restrictions under current COVID-19 safety regulations. It severely impacted travel, sampling, appointment of both graduate and post-doctorate workers. Some in-person seminars and webinars did take place before and after meeting restrictions. The target audiences are mostly grape and wine industry professionals. Although interactions with fellow scientists for peer-reviewed feedback are also undertaken. The PI's on this project also met with the

advisory board that represents fellow scientists, nurseries, vine testing facilities and grape and wine industry representatives in Oct and Nov of 2019. The next meeting will be scheduled after Thanksgiving 2020. Due to slower progress than expected because of several mitigating circumstances (COVID-19, natural disasters and more), the meeting has been delayed from its intended time of mid-November 2020 and will take place virtually. The PI, co-PI and collaborators met virtually in May 2020 to discuss progress, collaborations, and stumbling blocks.

Products

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Accepted	2020	NO

Citation

Bick, E.N., C.R. Preto, and F.G. Zalom. 2020. Timing the implementation of cultural practices for Spissistilus festinus (Hemiptera: Membracidae) in California vineyards using a stage-structured degree day model. J. Econ. Entomol. 113: 2558–2562.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Accepted	2020	NO

Citation

Levin A.D. and A. KC. 2020. Water Deficits Do Not Improve Fruit Quality in Grapevine Red Blotch Virus-Infected Grapevines (Vitis vinifera L.). Frontiers in Plant Science 11:1292. DOI: 10.3389/fpls.2020.01292.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Accepted	2020	NO

Citation

Wilson, H., Yazdani, A. S., and Daane, K. M. 2020. Influence of riparian habitat and ground covers on threecornered alfalfa hopper Spissistilus festinus (Hemiptera: Membracidae) populations in vineyards. Journal of Economic Entomology 113(5): 2354–2361. doi: 10.1093/jee/toaa151

Other Products

Product Type

Audio or Video

Description

A webinar recording is available on the UCD Viticulture and Enology website that describes the USDA-NIFA-SCRI grant, major goals, timelines, and achievements so far. As this is the first year of the grant, we expect outputs to increase exponentially in the coming years.

Changes/Problems

COVID-19 shelter-in-place and later travel and research safety measures at the different academic institutions have and had a significant impact on current milestones. The most important impact has been on new hires as it was extremely difficult to appoint new personnel especially with visa processing centers closed. Furthermore, access to field trials and laboratories for research were severely limited. Some objectives were majorly impacted and others marginally depending on the research activities needed and the individual personnel needs. However, progress have been made in most objectives on which we will build during the second year of funding.